## **UNCLASSIFIED**

AD NUMBER
AD840394
NEW LIMITATION CHANGE
TO Approved for public release, distribution unlimited
FROM Distribution authorized to U.S. Gov't. agencies and their contractors; Critical Technology; 30 JAN 1968. Other requests shall be referred to Department of the Army, Fort Detrick, MD.
AUTHORITY
SMUFD D/A ltr, 15 Feb 1972

DATE: 30 yanuary 192

### DDC AVAILABILITY NOTICE

Reproduction of this publication in whole or in part is prohibited. However, DDC is authorized to reproduce the publication for United States Government purposes.



#### STATEMENT #2 UNCLASSIFIED

This document is subject to special export controls and each transmittal to foreign governments or foreign nationals may be made only with prior approval of Dept. of Army, Fort Detrick, ATTN: Technical Release Branch/ TID, Frederick, Maryland 21701

#### SHEEP POX (ECTHYMA CONTAGIOSUM) IN CHAMOIS\*

Zentralblatt fuer Bakteriologie (Bacteriological Journal) Vol. CXCV, 1964, pages 175-181 W. Grausgruber

From the Bundesanstalt fuer Tierseuchenbekaempfung (Federal Animal Epidemic Institute), Moedling, Prof. F. Kress, Director.

Received on 1 October 1964

A disease generally isolated but occasionally epidemic has long been known in chamois; skin changes occur, especially in the lip area, and are accompanied by a frequently considerable crust and scab formation and can have a papillomatous appearance. The disease, generally called "papillomatosis of chamois," has other common names, such as "infectious papillomatosis," "contagious ringworm," "contagious papilloma," and "stomatitis papillomatosa." Many hunters speak of "winter mange of chamois" [9-11, 22].

The etiology long remained unknown. Not until 1951 did Bouvier, Burgisser, and Schweizer [3], who described in detail the phenomena of the disease in three chamois and an ibex, show that the chamois papillomatosis appeared, on the basis of the lesions, the disease's progress, and the complications, to be identical with ecthyma contagiosum of sheep. The Swiss authors made several transmission experiments, in addition. After scarifying two sheep on the thigh inner surface, they were able to infect them by using the skin crusts of the two chamois. It was also possible to infect a sheep and a goat with the ibex's crust material after light

\*Paper read before the Oesterreichische Gesellschaft fuer Mikrobiologie und Hygiene (Austrian Society of Microbiology and Hygiene) on 4 April 1964. scarification in the oral area.

An epidemic and unusually costly occurrence of this disease in chamois in the 1962/1963 winter in Austria, reported on elsewhere by Kress [13], initiated a series of experimental studies chiefly intended to contribute to further knowledge of the etiology and epizootiology.

At first, transmission experiments were made on sheep and goats. The transmission material came from chamois sent from various districts and representing variously severe sickness changes (Fig. 1). The crustally changed skin regions were removed and either simply grated into isotonic common salt solution or, if culturing tests were to be made simultaneously, pretreated with antibiotics (penicillin and streptomycin). The material was then rubbed into the lightly scarified skin of the lip region of the test animals.

In this way, it was possible in every instance to transmit the disease to sheep and goats. Within 3 to 4 days, small nodules first formed at the infection site, followed by vesicles and pustules, which often ran together and quickly dried, with crust formation of varied intensity. The crusts then dropped off, and, in favorable cases, by the 14th day after infection only slight swelling and reddening of the skin were evident at the infection site. In a few animals, it was 3 to 4 weeks before the disease phenomena had completely abated (Fig. 2).

For comparison, transmission experiments were made on sheep and goats, with the ecthyma contagiosum virus of sheep.) The characteristic phenomena of sheep pox formed in the experimental animals, infected by scarification in the lip area. The clinical picture here was exactly the same as for the sheep and goats which had been infected with crustal material derived from sick chamois.

Finally, ecthyma virus of ovine provenance was transmitted to three chamois in manner similar to that for the other test animals. In these animals, too, again starting on the fourth day after infection, the nodules, vesicles, and pustules, which rapidly became crusted over, formed at the scarification site. After the crusts fell off, 14 days after inoculation in one chamois and 21 and 30 days for the other two, only several minor thickenings of the skin were evident

The virus strain was kindly made available to us by Dr. B. Hess, Hanover.

at the scarification site. The fodder intake in these chamois, as in the other test animals, was unchanged even at the high point of the disease.

In addition to the transmission experiments on sheep, goats, and chamois, a series of culture experiments was made.

The various efforts to culture ecthyma virus from sheep or humans in chick embryoes has had little success as yet. Abdussalam [1] saw minor choricallantois membrane changes in some of the eggs inoculated with ovine virus; the changes were clearest 48 to 72 hrs after inoculation, but disappeared from the fourth passage on. Membranes of the fourth egg passage proved infectious for lambs, but those of the sixth passage were not. Although Lyell and Miles [16] found similar lesions of the chorionallantois membrane, Glover [6] was unable to show any changes after the second passage. In the extensive vesicular lesions of the choricallantois membranes, described by Borcila and Isopescu [2], nonspecific changes were undoubtedly involved, but the test of infectiousness led to results similar to those in the investigations of Abdussalam [1]. Hart, Hayston, and Keast [8], as well as MacDonald [17], were unable to see specific changes in the choricallantois membrane after inoculation with ecthyma virus. The culturing attempts made by Liess [14], showing membrane suspensions of even the second passage to be no longer infectious, gave negative results, too.

For cultures in chick embryoes, we used eggs hatched 12 days and we inoculated the chorioallantois membranes with virus-containing test material pretreated in the usual way with antibiotics. The eggs were then hatched 3 to 4 days more, the chorioallantois membranes were removed, and additional passages made. It was not possible to propagate the ecthyma virus of sheep in this way. Unsuccessful, too, were the attempts made with test material derived from chamois. The various changes observed in the chorioallantois membranes, such as thickenings or grayish white foci, appeared in the controls, as well. It was finally impossible to infect a lamb and a kid with chorioallantois membrane suspensions of the third egg passage. Both animals proved completely sensitive to a later infection with virus-bearing material from sheep or chamois.

Ecthyma virus from sheep and from humans has often been grown in tissue cultures, however. Thus, Webster [21] was able to propagate a smallpox virus strain in suspension cultures from sheep's skin tissue. Frederiks and Frenkel [4] used embryonal skin from sheep and cattle. Greig [7] successfully cultured ovine ecthyma virus in single-layer cell

cultures from embryonal sheep skin. Nagington and Whittle [18] isolated an ecthyma contagiosum virus strain from humans in amnion cell cultures, and Ramyar [20] isolated ovine virus in ape kidney cell cultures. Plowright, Witcomb, and Ferris [19] used sheep and calf testicular cell cultures for virus breeding, and Liess [14] was successful in culturing ecthyma virus of human and ovine origin in this manner and transmitting it in passages.

We used the same methods to continue the culture of ecthyma virus from sheep; young pig testicular cell cultures, too, proved suitable for this purpose.

In the same manner as in sheep pox, we were able to isolate and further culture in tissue cultures the virus from the infectious material derived from chamois. Inoculation of the cell cultures with the material from chamois produced exactly the same cytopathic changes as those caused by the ecthyma virus of cvine origin and previously described in detail by Liess [14]. The cytopathic changes became visible 24 hrs after inoculation with test material derived from chamois and led after 4 to 5 days to extensive destruction of the cell substrate. By reverse transmission of the individual tissue culture passages to sheep and goats, infectious virus could be demonstrated in every case.

Finally, to prove immunological identity, cross-immunisation experiments were made. Sheep and goats which had been infected with skin crust material from diseased chamois proved immune to an infection with ecthyma virus from sheep 2 to 3 months later. It was not possible, either, by using virus-containing material from chamois, to infect sheep and goats previously infected by ecthyma contagiosum virus of ovine origin. In two chamois previously infected with ecthyma virus from sheep it was not possible to initiate any disease phenomena by an inoculation done 3 months later from infectious skin crusts of diseased chamois.

The similarity of the clinical picture in chamois, sheep, and goats, the results of the transmission experiments, the uniform behavior in tissue cultures, the demonstration of cross-immunity, and finally the results of Gerstl's electron-microscope studies [5] have corroborated the assumption that the so-called "papillomatosis of chamois" is identical to the ecthyma contagiosum of sheep and goats. Our investigations have so far been unable to show any differences between the virus from chamois and the ecthyma virus of ovine origin. The various ecthyma virus strains isolated from chamois of various regions behaved alike, too.

According to all reports to date, sheep pox in chamois regularly appears only in the winter months and disappears quite rapidly in the spring when fresh fodder is again available for the wild animals [9, 10, 12]. A prerequisite for the occurrence of an infection is minor injuries on the lips, which can occur more often in winter. Several previous investigators pointed this out. The nature of the fodder probably plays an essential part here. The lignified plants which are often the only available nourishment in the snowy winters can readily cause injuries on the skin or mucous membranes and thus form an entry for the virus. Again, there is a possibility of injury, followed by infection, from the frozen fodder present during extremely snowfree winters.

In our animal experiments, grown sheep and goats, feeding normally, could be successfully infected only after previous light scarification. A contact infection, without this preceding scarification, was successful only with two lambs. These animals, kept in a stall section with two sheep and a goat, became ill on the 7th day after the artificially infected sheep and goat showed the first symptoms of sheep pox. The disease phenomena were severer and lasted longer in these lambs than in the cutaneously infected animals.

Repeated minor lacerations, such as can occur especially in cold weather from unsuitable fodder, are undoubtedly important to the severity of the clinical picture in chamois. The result is probably chiefly the papillomatous changes on the palate, mucous membrane of the cheeks, or the back of the tongue.

The influence of repeated traumatic effects were to be seen in our infection experiments, too. In diseased sheep and goats from whom material was continually taken for culture experiments and electron microscope studies, fresh efflues-cences always appeared, and the clinical picture was considerably more severe than in those animals whose infection was allowed to heal undisturbed (Fig. 3).

The prevailing view, that sheep pox occurs in chamois only in winter, appears to us not entirely tenable, for various reasons. In fact, we were able to find sheep pox during July and August in three chamois which had been shot for other reasons and sent for investigation. Two of these animals showed only very small encrusted areas in the lip region which would very easily have escaped observation in a less searching study and which could not have directly been suspected as ecthyma. In the third case, the crusted skin changes were somewhat clearer, and even the hunter noted it and suspected chamois mange. With all three chamois, it was possible to

prove, by virus isolation and transmission experiments with sheep, that sheep pox is involved in the skin changes. In one case it was additionally possible to show directly by electron microscope the ecthyma virus in the material taken from the chamois.

These investigations show that the appearance of sheep pox in chamois is not limited to the winter months but can occur in wild animals at any time of year, as would be expected. It is merely that in summertime, for the reasons mentioned, the cases are marer, the symptoms are considerably fainter, and the disease's progress milder, so that the diseased animals generally escape notice. It is additionally very probable that an infection with ecthyma virus is involved in at least part of the cases described by hunters as grasing animal mange."

Finally, the length of immunity in sheep pox was tested in a few animal experiments. It was found that sheep and goats once infected with both the ecthyma virus from ovine sources as well as the virus material proved resistant to a subsequent infection a year later with material from sheep or chamois. A chamois once infected with ecthyma virus from sheep was immune to an infection a year later with virus material from chamois.

#### SUMMARY

# Ecthyma contagiosum of chamois W. Grausgruber

Experimental investigations confirmed the identity of the conditions of "Chamois papillomatosis" and ecthyma contagiosum of sheep and goats. Like the ovinia derived ecthyma virus, the virus obtained from chamois could also be cultured in calf or pig testicular cell cultures. No differences have so far been demonstrated between the ovinia ecthyma contagiosum virus and virus strains derived from chamois.

The importance of minor injuries for establishing the infection and developing the disease changes also finds expression in animal experiments.

As opposed to present opinion, that ecthyma contagiosum only appears in chamois in winter, it is stressed that the disease may occur in game at any time of the year, even if it is less common and milder in form. This is confirmed by the demonstration of ecthyma virus in three chamois shot at the height of summer.

Sheep, goats and chamois infected with ecthyma contagiosum virus from sheep or chamois were found to be immune to reinfection one year later.

- Fig. 1. Sheep pox (ecthyma contagiosum) in a chamois.
- Fig. 2. Sheep, cutaneously infected with virus-containing skin crusts of a chamois. Ecthyma symptoms on 10th day after incoulation.
- Fig. 3. Goat (kid), cutaneously infected with ecthyma contagiosum virus (chamois strain). Clinical picture on the llth day after inoculation after repeated removal of material for study.

[ No bibliography given]

7